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(71) Applicant: THE PERKIN-ELMER CORPORATION [US/US]; 850 Lincoln Centre Drive, Foster City, CA 94404 (US).

(72) Inventor: NORDMAN, Eric, S.; 2150 Middlefield Road, Palo Alto, CA 94301 (US).

(74) Agents: FRAZIER, Jeffery, D. et al.; Dehlinger & Associates, P.O. Box 60850, Palo Alto, CA 94306-0850 (US).

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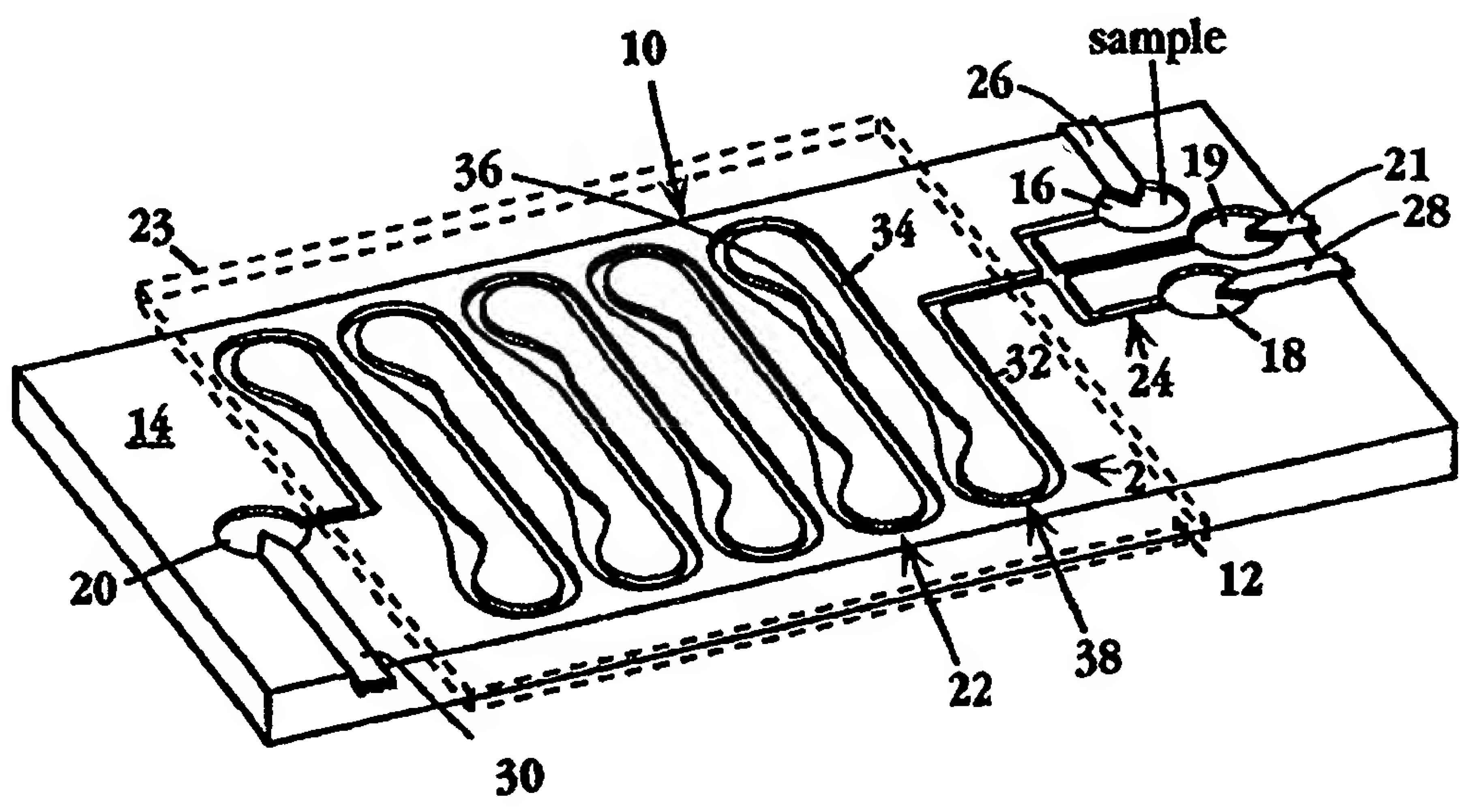

(57) Abstract

A serpentine electrophoresis channel, e.g., for a microchip format, is disclosed. The channel includes pairs of linear segments, e.g., parallel or right-angle segments, each joined by an angled channel region having a first curved channel portion subtending an angle $\alpha_f > \alpha$, where α is the angle between segments in a pair, and a second curved channel portion subtending an angle $\alpha_s = \alpha_f - \alpha$. The angles and cross sections of the two channel portions are such that δt_f, the time differential of analyte migration at inner and outer tracks in the first curved portion is equal to δt_s, the time differential of analyte migration at outer and inner tracks in the second curved portion, respectively.

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SERPENTINE ELECTROPHORESIS CHANNEL WITH SELF-CORRECTING BENDS

Field of the Invention

The present invention relates to electrophoretic separation devices, and in particular, to a device having a serpentine separation channel, for example, in a microfabricated device.

Background of the Invention

Electrophoresis exploits the differential rate of migration of charged species through a separation medium, under the influence of an electric field, for purposes of separating and/or characterizing physical properties of the charged species. Typically, the sample containing the charged species to be separated is placed at one end of a separation channel (which may be a linear channel or a lane in a 2-dimensional slab) and a voltage difference is placed across opposite channel ends until a desired migration end point is reached. The separated analyte molecules may then be detected, *e.g.*, by optical detection, radiography, or band elution.

As examples, gel electrophoresis in the presence of a charged surfactant, such as dodecyl sulfate, is widely used for protein separation and for characterizing protein molecular weight. Electrophoresis in a gel or liquid medium is commonly used to separate oligonucleotides with different numbers of bases, for example, in DNA sequencing.

One of the possible applications of microfabrication techniques that has been proposed is in the area of column separation devices, including electrophoresis devices. Jacobsen, *et al.* (*Anal. Chem.* 66:2369 (1994); *Electrophoresis* 16:481 (1995) have described a "microchip" electrophoresis device formed by etching an open electrophoresis channel, and suitable connecting reservoirs, on a glass slide. Because of the small chip dimensions, typically less than 10-15 cm on a side, it is necessary to form the separation column in the form of a serpentine pathway in order to achieve total column separation lengths suitable for most applications.

Although a serpentine column solves the problem of adequate column length on a microchip, it introduces a potentially serious limitation in terms of column resolution. When a electrophoretic band is migrating through a linear channel, the molecules making up the band, which are all migrating at roughly the same speed, tend to migrate as a tight band. However, the same molecules migrating through a turn in a serpentine pathway will migrate through the shorter inner side of the channel faster than through the longer outer side of the channel, leading to band spreading and nonuniformity across the width of the channel. At each turn in the pathway, more band resolution is lost. Heretofore, this problem has severely limited the

range of practical electrophoresis applications in a microchip format.

Summary of the Invention

The application includes, in one aspect, an electrophoresis channel through which one or more charged species are intended to migrate under the influence of a voltage difference placed across opposite ends of the channel. The channel includes (i) a pair of channel segments disposed at an angle α with respect to one another, and (ii) an angled channel region connecting the two channel segments.

The angled channel region has a first curved channel portion subtending an angle $\alpha_f > \alpha$, where α is the angle between the two channel segments, and a second curved channel portion subtending an angle $\alpha_s = \alpha_f - \alpha$. The first curved portion defines inner and outer tracks or channel sides, such that an analyte migrating through the first channel portion under the influence of such voltage difference will traverse the inner track in a time interval δt_f faster than that of the same analyte traversing the outer track. The second curved portion defines second inner and outer tracks such that an analyte migrating through the second channel portion under the influence of the same voltage difference will traverse the outer track in a time interval δt_s faster than that of the same analyte traversing the outer track. The angles and cross-sections of the two channel portions are such that δt_f is approximately equal to δt_s.

The channel is typically part of a serpentine pathway containing a plurality of such segments, each pair of adjacent channel segments being connected by an associated angled channel region.

Where the two channel segments are disposed at right angles with respect to one another, α_f is preferably between about 110° and 160°, and α_s, between about 20° and 70°, respectively. Where the two channel segments are disposed substantially parallel to one another, α_f is preferably between about 200° and 250°, and α_s, between about 20° and 70°, respectively.

In a microfabricated chip format, the channel has preferred width dimensions between about 25-250 microns, and preferred depth dimensions between about 5-100 microns.

In one general embodiment, the first and second curved portions have substantially constant channel widths W_f and W_s, respectively, where $W_f < W_s$. In this embodiment, the angled channel region further includes tapered-width segments joining the second curved channel portion to the first channel portion and to one of the two channel segments. An approximate relationship between W_f and W_s is given by the relationship $W_s = (\alpha_f W_f^2 R_f / \alpha_s R_s)^{1/2}$, where R_f and R_s are the radii of curvature of the first and second curved portions, respectively.

In another general embodiment, the first curved channel portion has a preferably fixed channel width, and the second channel portion, a variable width that expands on progressing inwardly from each end.

In yet another embodiment, the first curved channel portion has a channel depth which increases on progressing toward the second channel portion, and the second curved channel portion has a channel depth which decreases on progressing away from the first curved channel portion. The channel width may be substantially constant in the channel segments and the channel connecting region therebetween.

More generally, the invention includes an analyte separation channel through which one or more analytes is intended to migrate under the influence of a motive force applied to opposite ends of the channel. The device includes (i) a pair of channel segments disposed at an angle α with respect to one another, and (ii) an angled channel region of the type just described connecting the two channel segments. The motive force may be a voltage difference applied across the opposite ends of the channel, or a force producing fluid movement through the channel or a combination of the two.

In a related aspect, the invention includes a microfabricated device for electrophoretic separation of analytes in a mixture. The device includes a substantially planar-surface substrate having formed thereon, first and second reservoirs and a serpentine electrophoretic channel extending therebetween. The channel has a plurality of linear segments, and connecting the adjacent ends of each pair of adjacent segments, an angled channel region of the type described above. The channel, including the linear segments and angled channel regions, has preferred channel width dimensions between about 25-250 microns, and depth dimensions between about 5-100 microns.

These and other objects and features of the invention will become more fully apparent when the following detailed description of the invention is read in conjunction with the accompanying drawings.

Brief Description of the Drawings

Fig. 1 is a perspective view of a microfabricated device constructed according to the present invention, having an open electrophoresis channel and liquid reservoirs formed on a substrate;

Fig. 2 is an enlarged view of a 180° bend in a serpentine channel formed in accordance with the present invention, illustrating the effect of the self-correcting bend on band distortion;

Figs. 3A-3C are sectional views taken along lines 3A-3A, 3B-3B, and 3C-3C,

respectively, in Fig. 2;

Fig. 4 is an enlarged view of a 90° bend in a serpentine channel formed in accordance with one embodiment of the present invention;

Fig. 5 is an enlarged view of a 90° bend in a serpentine channel formed in accordance with another embodiment of the invention;

Fig. 6 is an enlarged view of a 90° bend in a serpentine channel formed in accordance with yet another embodiment of the invention; and

Fig 7 is a cross-section of the channel region in Fig. 6, taken along the channel pathway 7-7 in Fig. 6.

Detailed Description of the Invention

Fig. 1 shows a microfabricated device 10 constructed in accordance with the invention, for electrophoretic separation and/or characterization of one or more analytes in a sample mixture. The device generally includes a planar substrate 12 having formed in its upper surface 14, open reservoirs 16, 18, 19, and 20, and a serpentine electrophoresis channel 22 connecting the reservoirs. Reservoirs 16 and 18, which are intended to contain electrophoresis buffer and sample fluid, respectively, are connected in fluid communication with each other and with channel 22 through a fork-like connector 24. Reservoirs 19, 20 are intended to hold the waste reservoir. The four reservoirs are connected to electrodes 26, 28, 21, and 30, as shown, which are in turn connected to suitable voltage leads during operation of the device, for (i) loading sample from reservoir 16 into channel 22, by applying a voltage across electrodes 26, 28, and (ii) (ii) electrophoretically separating charged sample components, by applying a voltage difference across opposite ends of the channel, i.e., across electrodes 21, 30.

With continued reference to Fig. 1, channel 22 includes a plurality of parallel linear channel segments, such as segments 32, 34, and 36, and curved channel regions connecting the adjacent ends of adjacent linear segments, such as curved channel region 38 connecting adjacent ends of segments 32, 34. In a typical embodiment, the substrate or chip has side dimensions of between about 1 to 15 cm, and the linear segments are each about .5 to 10 cm in length. Thus, for example, a channel having 30 linear segments, each about 8 mm in length has a column length, ignoring the lengths of the connecting regions, of about 250 mm. With the added lengths of the connecting regions, the total length may be in the 30 cm range, on a chip whose side dimensions may be as little as 1 cm. A coverslip 23 placed over the portion of the substrate having the serpentine channel serves to enclose the channel, although an open

serpentine channel is also contemplated.

The construction of a curved connecting region-- in this case, region 38-- is shown in enlarged plan view in Fig. 2, which shows portions of linear segments 32, 34 connected by the region. The region includes a first curved channel portion 40 which subtends an angle α_f which is greater than the minimum angle α needed to connect the two segments. Where, as here, the linear segments are parallel and α is 180°, α_f is typically between about 200°-250°, *i.e.*, about 20°-70° over the minimum angle. As shown, portion 40 has a substantially constant channel width W_f along its length, equal to the channel width of the connected linear segments.

As seen in Fig. 3A, which is a cross-section along line 3A-3A in Fig. 2, the channel has a substantially rectangular cross-section with a width dimension W_f and depth dimension d_f. W_f is typically between about 25-200, preferably 50-100 microns, and d_f is typically about 5-100, preferably 25-75 microns.

With continued reference to Fig. 2. portion 38 includes a second curved channel portion 42 subtending an angle α_s which corrects the overangle α_f to provided the desired 180° total angle in the curved portion; that is, $\alpha_s=\alpha_f-\alpha$. Thus, for example, where α is 180°, and α_f is between about 210° and 250°, α_s is between about 20° and 70°, respectively. The width W_s of the second curved portion is greater than W_f and is selected, in relation to the two angles α_f and α_s, and in accordance with the invention, to correct band distortion produced as a band moves through portion 40, as will be described below. In the embodiment illustrated, and as shown in Fig. 3C, W_s is greater than W_f, acting in effect reduce the electric field strength on analyte molecules migrating through this portion, relative to portion 40. As seen in Fig. 3C, the channel depth d_s in portion 42 is the same as that in portion 40, *i.e.*, $d_s=d_f$.

Channel region 38 further includes two tapered-width segments 44, 46, which serve as interfaces between (i) the smaller-width portion 40 and the larger-width portion 42 (segment 44) and between (ii) the larger-width portion 42 and the smaller-width linear segment 34 (segment 46). A cross-sectional view of segment 44 is shown in Fig. 3B, showing a channel width intermediate between that of portions 40, 42, and the same channel depth.

The operation of the second channel portion, in correcting curved channel effects produced in the first channel portion, will now be discussed, also with reference to Fig. 2. In this figure, a charged species migrating as a band through the channel is indicated at various stages through the curved channel regions by numerals 48a-48g. Band 48a, which is at the position just entering the curved channel portion, is substantially undistorted, meaning that the band is both narrow and disposed along an axis substantially normal to the channel axis. As

the band enters channel portion 40, it begins to distort, as shown at 48b, due to the shorter migration distance of molecules along the inner track 40a and the longer migration distance of molecules along the outer track 40b. The distortion increases progressively as the band migrates through portion 40, as illustrated by bands 48c and 48d.

It can be shown that a band on the inside track will lead a band on the outside track with a time δ_f approximately equal to $\alpha_f(2W_fR_f)/\mu E_{f\text{-center}}$, where R_f is the radius of curvature of curved portion 40, W_f is the channel width, μ is the mobility of the migrating species, in $m^2/Vsec$, and $E_{f\text{-center}}$ is the electric field in portion 40 at the center of the track, resulting from the potential difference applied across opposite ends of the channel.

The purpose of the second curved portion is to provide a correction, on the opposite channel side, for the band distortion produced in the first curved portion. Briefly, this second curved portion is designed such that a band on the outside track 42b (which is now the shorter of the two tracks) will lead the band on the inside track 42a by a time δ_s substantially equal to δ_f. Similar to the calculation above, it can be shown that δ_s is approximately equal to $\alpha_s(2W_sR_s)/\mu E_{f\text{-center}}$, where R_s is the radius of curvature of curved portion 42, W_f is the channel width, μ is the mobility of the migrating species, in $m^2/Vsec$, and $E_{f\text{-center}}$ is the electric field in portion 42 at the center of the track, also due to the same potential difference applied across the ends of the channel. It is noted that $E_{s\text{-center}}$ is less than $E_{f\text{-center}}$, due to the larger channel width in channel 42, according to the relationship $E_s=E_f(W_f/W_s)$. The condition $\delta_f=\delta_s$ is satisfied when $\alpha_f(2W_fR_f)/\mu E_{f\text{-center}} = \alpha_s(2W_sR_s)/\mu E_{f\text{-center}}$, that is, when $\alpha_f/\alpha_s=W_s^2R_s/W_f^2R_f$. As an example, assume W_s is 50 μm, α_s is 210° α_f is 30°, and $R_f=R_s=1$ mm. W_s is then $((50\ \mu m)^2(210/30))^{1/2}$, or about 132 μm.

With reference again to Fig. 2, it can be appreciated that band 48d migrates through tapered segment 44 substantially without correction, is fully corrected within portion 42, and then migrates through segment 46 and into segment 34 in corrected form, *i.e.*, with the band axis oriented substantially normal to the segment axis.

Fig. 4 shows an embodiment of a 90° curved channel region 50 constructed in accordance with the invention, for use, for example, in a serpentine channel of the type described above, but where each 180° turn is produced by two adjoining 90° turns. Channel region 50 joins two linear channel segments 52, 54 which in this embodiment are disposed at right angles with respect to one another.

Channel region 50 includes a first curved channel portion 56 which subtends an angle α_f which is greater than 90°, and a second channel portion 58 which subtends an angle α_s which corrects the overangle α_f to provide the desired 90° total angle in the curved portion;

that is, $\alpha_s=\alpha_f-\alpha$. In the α=90 embodiment, α_f is typically between about 110° and 160°, and α_s is between about 20° and 70°, respectively. As in the 180° embodiment, the width W_s of the second curved portion is greater than W_f and is selected, in relation to the two angles α_f and α_s, and in accordance with the invention, to correct band distortion produced as a band moves through portion 56, as will be described below. In the embodiment illustrated, where the channel depth is uniform throughout the channel region W_s is greater than W_f, and related through the relationship $\alpha_f(2W_fR_f)/\mu E_{f\text{-center}} = \alpha_s(2W_sR_s)/\mu E_{f\text{-center}}$, or equivalently, when $\alpha_f/\alpha_s=W_s^2R_s/W_f^2R_f$, where R_f, R_s, μ, $E_{f\text{-center}}$ and $E_{f\text{-center}}$ are as above. As an example, assume W_s is 50 µm, α_s is 120° and α_f is 30°, and R_f=R_s=1 mm. W_s is then (50 µm^2(120/30))$^{1/2}$, or 100 µm.

Region 50 further includes tapered segments 60, 62 which serve as interfaces between (i) the smaller-width portion 56 and the larger-width portion 58, and (ii) the larger-width portion 42 and the smaller-width linear segment 54.

Analogous to the band behavior in the 180° turn region 38, an analyte band migrating into portion 56, substantially normal to the axis of segment 52, will become distorted by its migration through portion 56, with the outer-side of the band trailing the inner side of the band. The analyte migrates through tapered segment 60 substantially without correction, is fully corrected within portion 58 and then migrates through segment 62 and into segment 54 in corrected form, *i.e.*, with the band axis oriented substantially normal to the segment axis.

Fig. 5 shows another embodiment of a 90° curved channel region 64 constructed in accordance with the invention, for use, for example, in a serpentine channel 66 of the type described above. Channel region 64 joins two linear channel segments 68, 70 which in this embodiment are disposed at right angles with respect to one another. Channel region 50 includes a first curved channel portion 72 subtending an angle α_f which is greater than 90°, and a second channel portion 74 subtending an angle α_s which corrects the overangle α_f to provided the desired 90° total angle in the curved portion.

This embodiment differs from the one illustrated in Fig. 4 in that curved portion 74 replaces portion 58 and the two tapered segments 60, 62 in portion 50, as a continuously curved portion. That is, W_s is continuously variable through portion 74, from a minimum width W_s to a maximum width $W_{s\text{-max}}$. Exemplary angles α_s, α_f, are as above, where the radius of curvature R_s of portion 74 is about 3-4 times that in the Fig. 4 embodiment, but the angle α_s subtending the portion is about the same in both embodiments. The relationship between W_s and W_f is more complex than that shown above, but can be determined from the relationships given above, by integrating over α_s, where the value of W_s varies continuously over portion 74

according to a known angle-dependent relationship.

The operation of portion 74 in correcting band distortion produced in portion 72 is substantially as described above, but where band correction occurs over the entire region between portion 72 and segment 70.

Still another embodiment of the invention, for a 90° turn, is illustrated by angle channel region 76 in Fig. 6. The channel region, which joins right-angle channel segments 78, 80, includes a first curved channel portion 82 subtending an angle α_f>90°, and a second curved channel portion 84 subtending an angle α_s, which corrects the overangle α_f to provided the desired 90° total angle in the curved portion. Also forming part of the channel region are interface segments 86, 88 connecting portion 82 to portion 84, and portion 84 to segment 80, respectively. Exemplary α_f and α_s are as above.

The embodiment differs from those above in that the width W_f of portion 84 is the same the width W_s of portion 82, but portion 84 has a depth d_s which is greater than d_f, as illustrated in Fig. 7, which shows a segmented cross-section (through segments indicated by A, B, C, D, and E) along indicated portions of region 76. Also as seen, interface segments 86, 88 have tapered channel depths, rather than the tapered channel widths of the interface segments in the earlier described embodiments.

The electric field E_s in portion 84 is equal to $E_f(d_s/d_f)$, and band correction ($\delta t_f=\delta t_s$) occurs when $\alpha_f(2W_fR_f)/\mu E_{f\text{-center}} = \alpha_s(2W_sR_s)/\mu E_{s\text{-center}}$, that is, when $\alpha_f/\alpha_s=W_sd_sR_s/W_fd_fR_f=d_sR_s/d_fR_f$. As an example, assume d_s is 50 µm, α_s is 120° and α_f is 30°, and $R_f=R_s$=1 mm. W_s is then 50 µm (120/30), or about 200 µm.

The operation of region 76 in correcting band distortion is similar to that described above, for example, with respect to the embodiment shown in Fig. 4. Briefly, a band becomes distorted by its migration through portion 82, with the outer-side of the band trailing the inner side of the band. The band migrates through tapered segment 86 substantially without correction, is fully corrected within portion 84 and then migrates through segment 88 and into segment 80 in corrected form, *i.e.*, with the band axis oriented substantially normal to the segment axis.

From the foregoing, it can be appreciated how various objects and features of the invention are met. The invention is compatible with tightly coiled serpentine electrophoresis or other chromatographic channel configurations formed in a small-area microchip, for example, using conventional microfabrication techniques. The microfabrication method may involve either same-depth, variable-width etching, or same-width, variable depth etching, or a combination of the two.

The self-correcting bend feature of the invention acts to correct distortion produced by band migration around a turn, due to slower migration at the outside of the turn, acting to preserve band resolution along the entire channel length, which may include numerous turns, typically 90° or 180° turns.

Although the invention has been described with respect to specific embodiments, it will be appreciated that a variety of modifications may be made within the scope of the claimed invention. For example, the serpentine channel may be formed by chemical or laser etching techniques on a relatively large-scale plate, *e.g.*, a 10 cm × 10 cm plate designed for preparative electrophoresis or chromatography. The serpentine channel may be formed in a closed tube, such as a capillary electrophoresis tube, where each turn in the tube includes an expanded diameter, self-correcting counter turn. In still another aspect, the self-correcting turn may apply to other types of chromatography channels or tubes, dependent on pressurized fluid flow or gravity rather than a voltage difference as a motive force for moving analyte molecules through a separation medium.

IT IS CLAIMED:

1. An analyte separation device having a substrate in which is formed a channel through which one or more analytes are intended to migrate under the influence of a motive force across the channel, said channel comprising:

(i) a pair of channel segments disposed at an angle α with respect to one another, and

(ii) an angled channel region connecting the two channel segments, said region having

(a) a first curved channel portion subtending an angle $\alpha_f>\alpha$, and defining first inner and outer tracks such that an analyte migrating through the first channel portion under the influence of such force will traverse the inner track in a time interval δt_f faster than that of the same analyte traversing the outer track, and

(b) a second curved channel portion subtending an angle $\alpha_s = \alpha_f - \alpha$, and defining second inner and outer tracks such that an analyte migrating through the second channel portion under the influence of the same force will traverse the outer track in a time interval δt_s faster than that of the same analyte traversing the outer track,

where the cross-sections of said curved channel portions are such that δt_f is approximately equal to δt_s.

2. The device of claim 1, wherein the two channel segments are disposed at right angles with respect to one another, α_f is between about 110° and 160°, and α_s is between about 20° and 70°, respectively.

3. The device of claim 1, wherein the channel segments are disposed substantially parallel to one another, α_f is between about 200° and 250°, and α_s is between about 20° and 70°, respectively.

4. The device of any one of claims 1 to 3 which is formed in a microfabricated chip, and has channel width dimensions between about 25-250 microns, and depth dimensions between about 5-100 microns.

5. The device of any one of claims 1 to 4, which is part of a serpentine pathway containing a plurality of such segments, each segment pair connected by said angled channel region.

6. The device of any one of claims 1 to 5, wherein the first and second curved portions

have substantially constant channel widths W_f and W_s, respectively, where $W_f<W_s$, and the angled channel region further includes tapered-width segments joining the second curved channel portion to the first channel portion and to one of the two associated channel segments.

7. The device of claim 6, wherein $W_s=(\alpha_f W_f^2 R_f/\alpha_s R_s)^{1/2}$, where R_f and R_s are the radii of curvature of the first and second curved portions, respectively.

8. The device of any one of claims 1 to 5, wherein the first curved channel portion has a fixed channel width, and the second channel portion, a variable width that expands on progressing inwardly from each end.

9. The device of any one of claims 1 to 5, wherein the first curved channel portion has a channel depth which increases on progressing toward the second channel portion, and the second curved channel portion has a channel depth which decreases on progressing away from the first curved channel portion.

10. The device of claim 9, whose channel width is substantially constant in the channel segments and the channel connecting regions therebetween.

11. The device of any one of claims 1 to 10 for use in the electrophoretic separation of analytes in a mixture, wherein said motive force is a voltage difference applied across opposite ends of the channel.

12. A microfabricated device for electrophoretic separation of analytes in a mixture, comprising

a substantially planar-surface substrate having formed thereon, (i) first and second reservoirs and (ii) a channel, as defined in any one of the preceding claims, extending between said first and second reservoirs.

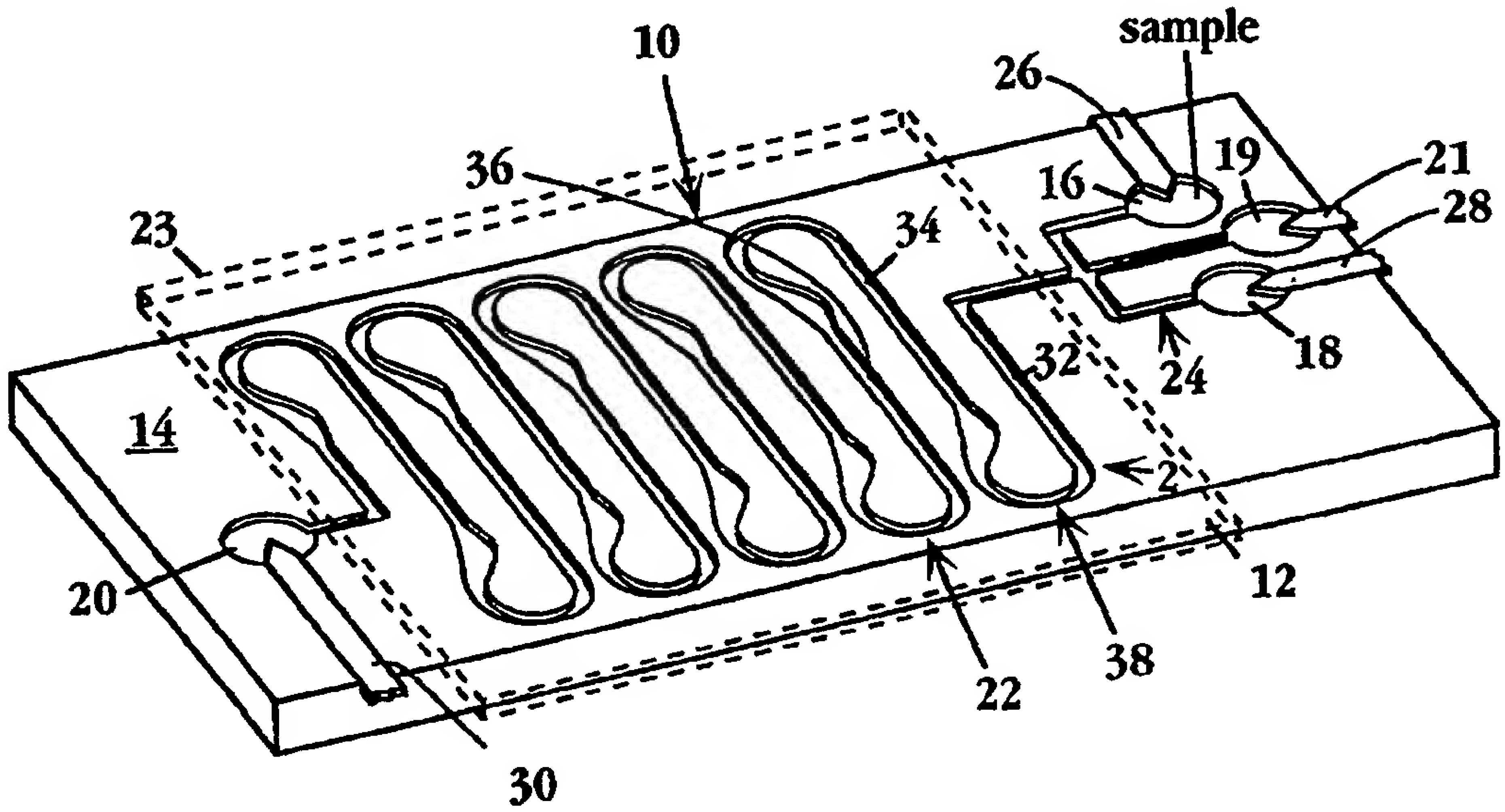
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Fig. 1

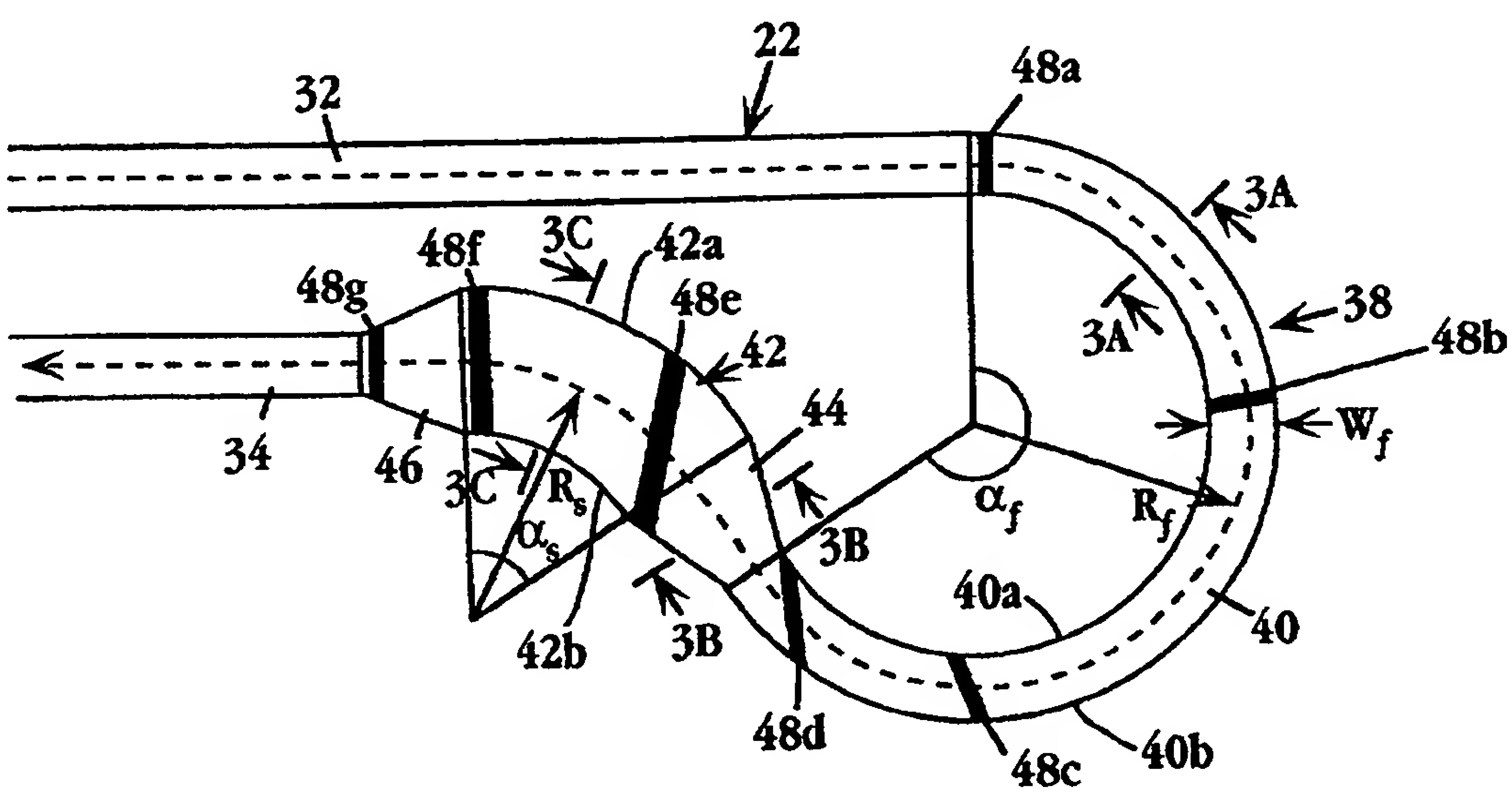
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Fig. 2

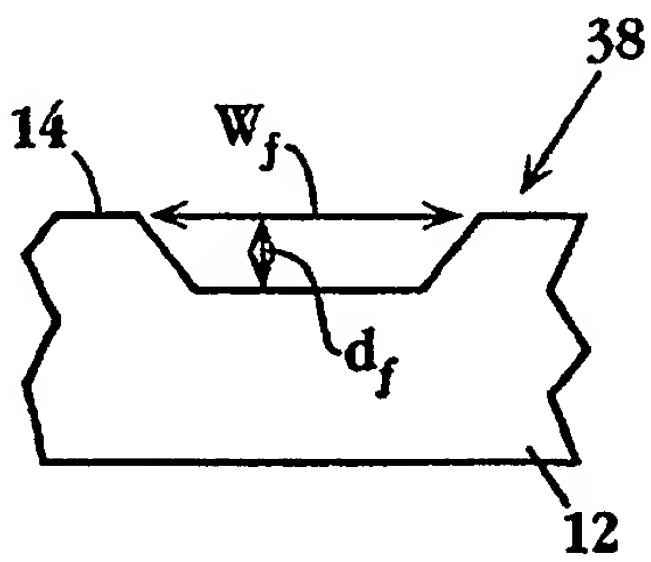


Fig. 3A

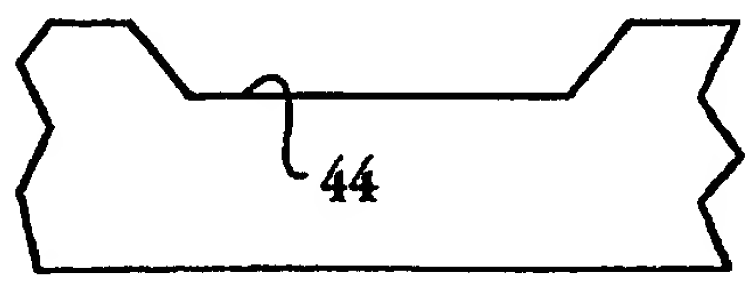


Fig. 3B

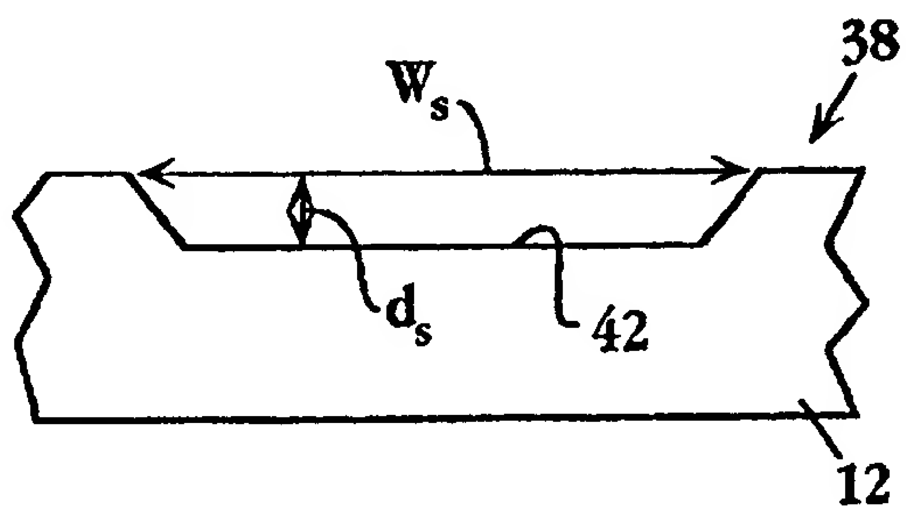


Fig. 3C

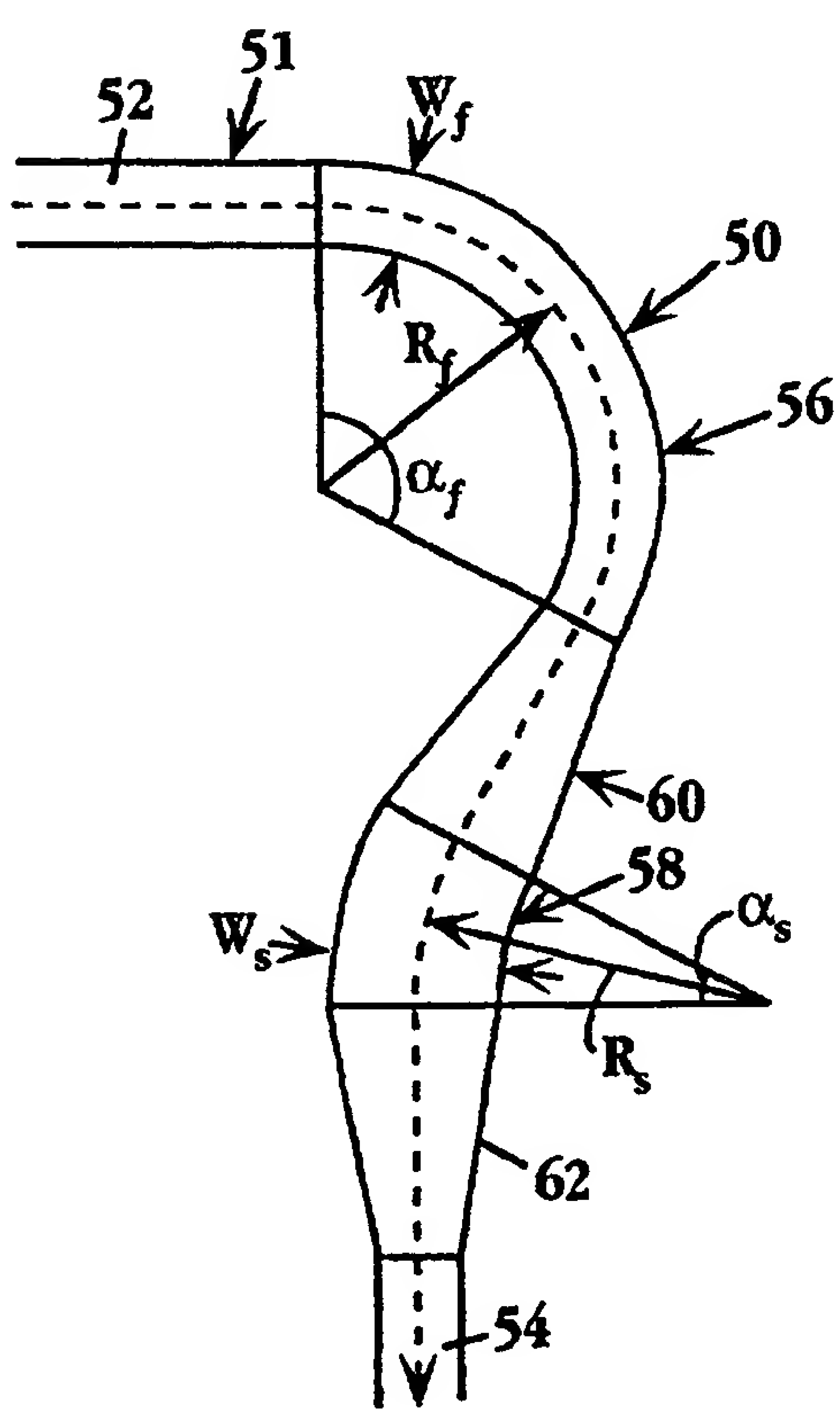


Fig. 4

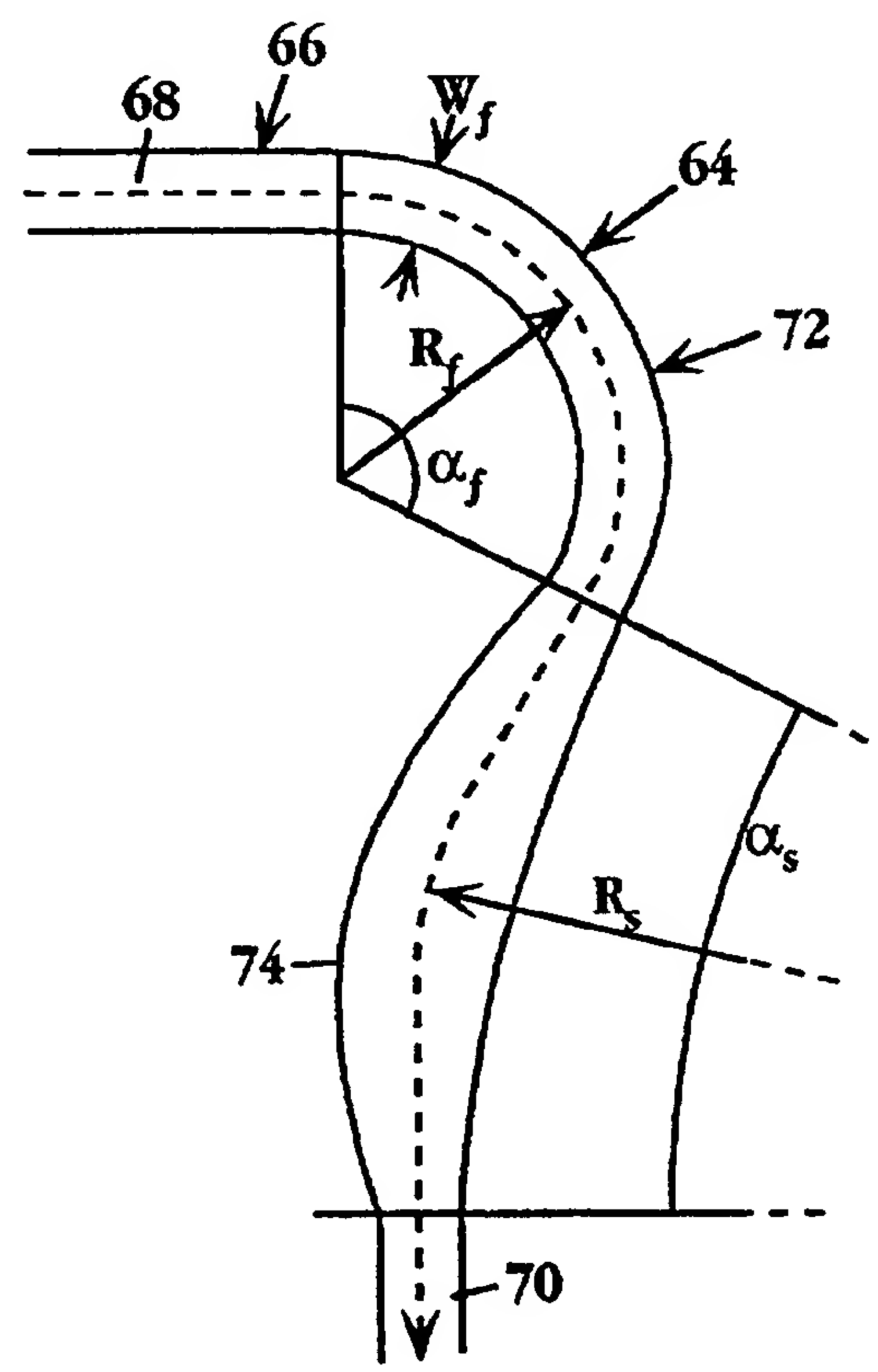
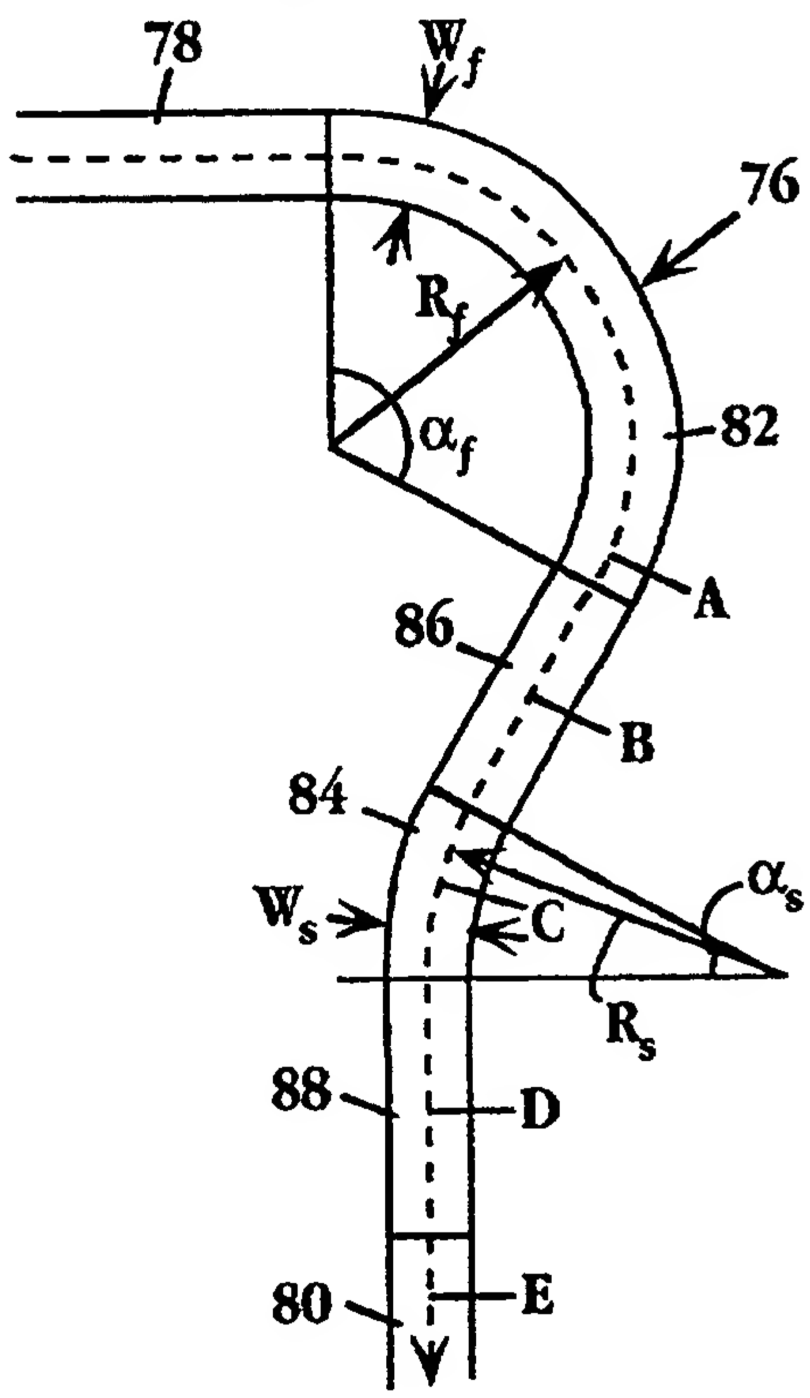


Fig. 5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R_f
α_f
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A
86
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α_s
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C
R_s
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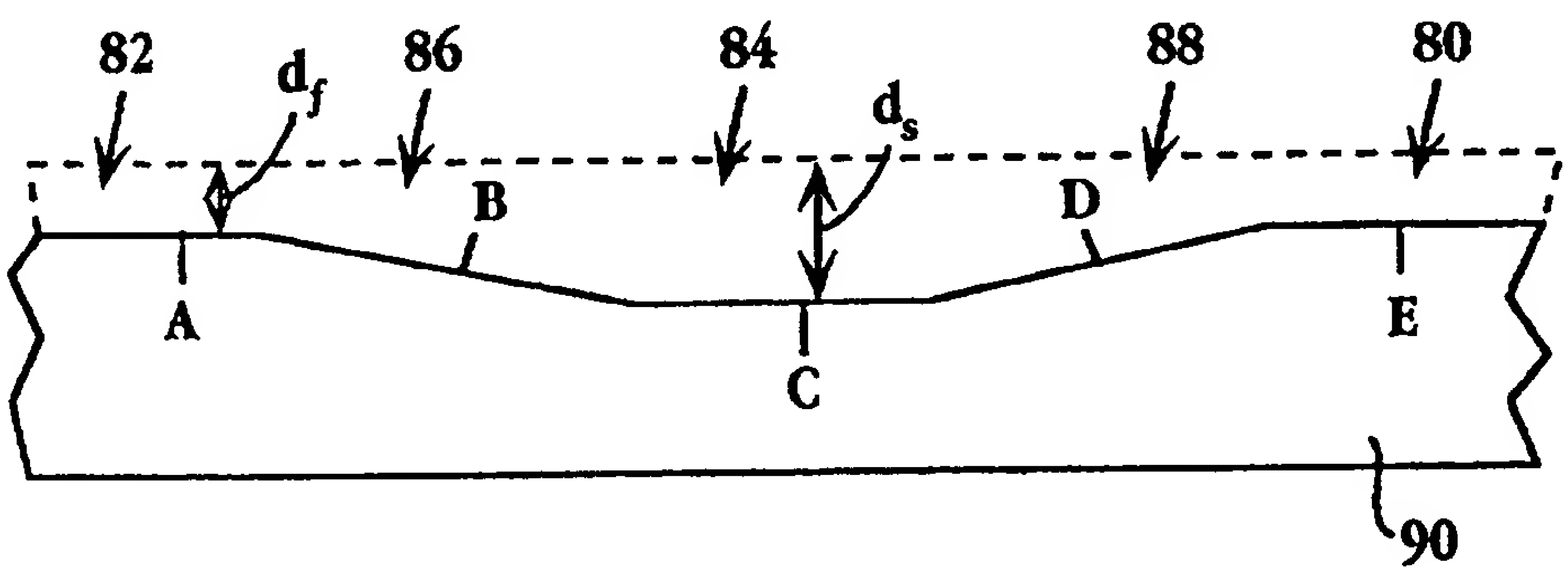
D
80
E

Fig. 6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B
D
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C
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90

Fig. 7

INTERNATIONAL SEARCH REPORT

In tional Application No
PCT/US 98/24202

A. CLASSIFICATION OF SUBJECT MATTER
IPC 6 G01N27/447

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 6 G01N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO 96 04547 A (LOCKHEED MARTIN ENERGY SYS INC ;RAMSEY J MICHAEL (US)) 15 February 1996 see figure 12	1,12
A	WO 91 16966 A (PHARMACIA BIOSENSOR AB) 14 November 1991 see figure 1	1

[ ] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

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"A" document defining the general state of the art which is not considered to be of particular relevance

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"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

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"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
19 February 1999	25/02/1999

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Duchatellier, M

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INTERNATIONAL SEARCH REPORT

Information on patent family members

Inte[illegible]ional Application No

PCT/US 98/24202

Patent document cited in search report		Publication date	Patent family member(s)		Publication date
WO 9604547	A	15-02-1996	AU	701348 B	28-01-1999
			AU	3150895 A	04-03-1996
			CA	2196429 A	15-02-1996
			CN	1168720 A	24-12-1997
			EP	0775306 A	28-05-1997
			JP	10507516 T	21-07-1998
			US	5858195 A	12-01-1999
WO 9116966	A	14-11-1991	SE	470347 B	31-01-1994
			AT	130528 T	15-12-1995
			DE	69114838 D	04-01-1996
			DE	69114838 T	05-06-1996
			EP	0527905 A	24-02-1993
			SE	9001699 A	11-11-1991
			US	5376252 A	27-12-1994